

## Crystal Structure of Colicin E3: An Enzymatic Cytotoxin

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Abstract No. Soel0440

Beamline(s): X25, X8C

**Introduction:** Colicins are toxic *E. coli* proteins composed of a translocation domain (T), a receptor-binding domain (R) and a catalytic or channel-forming domain (C). Colicin E3 acts as a specific ribonuclease that cleaves the phosphodiester linkage between bases 1493 and 1494 of 16S ribosomal RNA. This nick inactivates the ribosome and the cell dies. The producing organism has a mechanism to protect its ribosomes in the form of a specific inhibitor, the immunity protein, which forms a tight 1:1 complex with colicin E3.

**Methods and Materials:** The crystal structure was solved by MIR using five mercury and osmium derivatives. **Results:** The crystal structure of a binary complex of colicin E3 with its immunity protein has been determined to a resolution of 3Å. The structure is made up of the globular T and C domains, and an elongated intervening R domain which consists of a 100Å long helical hairpin. The T domain is a  $\beta$ -barrel and the small C domain forms a six-stranded antiparallel  $\beta$ -sheet. The immunity protein forms a wedge in between the C and T domains, and it makes contacts with both these domains. The immunity protein consists of a four-stranded antiparallel  $\beta$ -sheet flanked by three  $\alpha$ -helices on one side of the sheet, and it is very similar to the structure of free immunity protein determined earlier.

**Conclusions:** The length of the complex, 135Å, is long enough to span the outer membrane as well as the periplasm.

**Acknowledgments:** This project is funded by grant MCB97-28420 from the National Science Foundation